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SCIENTIFIC RESEARCH ON PAPAVER BRACTEATUM[1/]

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DETERMINATION OF THEBAINE IN PAPAVER BRACTEATUM[2/]

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1/ General note: The views expressed by collaborating scientists in their contributions to the programme are not necessarily shared by the Secretariat.

2/ In the report of the Second Working Group on Papaver bracteatum (document ST/SOA/SER.J/2), it was recommended that methods for the determination of thebaine in Papaver bracteatum should be compared and evaluated; to achieve this, it was decided that participants in the Working Group would forward their methods for publication in this series so that these could be tried out in the various laboratories.

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DETERMINATION OF THEBAINE IN PAPAVER BRACTEATUM

At the first Working Group in December 1972, I reported on the determination of the thebaine content of Papaver bracteatum. The analyses were carried out using the Sakurai method[3] as modified by B. Nielsen and J. Stene.[4]

Since that time, we have determined the thebaine content of the United Nations reference sample of P. bracteatum (UNB 6) and also of plant material raised in Norway from the seeds of P. bracteatum (UNB 2). Comparative analyses were carried out using the method of Fairbairn and Hakim and the method of Sakurai, as modified by Nielsen and Stene, and the results are given in Table 1.

The method of Fairbairn and Hakim was used in its original form as described by the authors in J. Pharm. Pharmacol., (1973), 25, 353-358. The Nielsen and Stene modification of Sakurai's method is given below.

Procedure

Mix 1 g of finely powdered sample with about 25 g of aluminium oxide (standardized activity II-III, Merck) and 2 ml of 15% sodium hydroxide solution. Transfer the uniform mixture quantitatively to a 18 x 220 mm chromatographic column. Elute the thebaine base with 200 ml of chloroform, at a rate not exceeding 2 ml per minute. Collect the eluate in a 500 ml round bottom flask and evaporate in vacuum to about 10 ml. Add 60 ml of 3.5% hydrochloric acid and heat and swirl until the chloroform is completely evaporated. 5 ml of the sample solution is placed in a 25 ml measuring flask and warmed in a boiling water bath for 25 minutes. When cooled, 4 ml of 0.5% 4-aminophenazone solution and 1.5 ml of 1% potassium ferricyanide solution are added and the mixture is shaken. After exactly one minute, 2 ml of 10% ammonia water is added, shaken again and warmed in a boiling water bath for 5 minutes.

The mixture is cooled with tap water to room temperature, diluted with 2.5 ml of ethanol and water to 25 ml. The absorbance of the solution is measured - after filtering - at 435 mμ.

3/ Sakurai, H., J. Pharm. Soc. Japan, (1960), 80, 909

4/ Of Weiders Pharmaceutical Company, Hausmannsgt.6, Oslo-1

Standard solution: Prepare a solution of pure thebaine base in 3.5% hydrochloric acid to give a concentration of 0.20 - 0.25 mg per ml. 5 ml of this solution is warmed for 25 minutes and the reagents added, as described above.

The blank test is carried out with the same reagents and in the same manner, using 5 ml of dilute hydrochloric acid instead of the test solution.

Table 1

Samples	Thebaine content in percentage, calculated on a dry weight basis according to:	
	Sakurai method as modified by Nielsen and Stene	Fairbairn - Hakim method
UN reference sample (powdered capsules) UNB 6	1.26	1.39
P. bracteatum cultivated in Norway from UNB 2 seeds		
Root	0.76 (0.39) x/	0.79
Leaves	0.80 (0,18) x/	0.81
Stem	0.58	0.77
Capsules	1.27	1.39

x/ The figures in parenthesis show the percentage of thebaine found in the young plants in the fall 1972.